

Dormancy and germination of Johnson grass seed (*Sorghum halepense* (L.) Pers.)

Dormantnost i klijavost sjemena divljeg sirka (*Sorghum halepense* (L.) Pers.)

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Abstract

The aim of the research was to determine the effectiveness of various methods for breaking seed dormancy of weed species Johnson grass (*Sorghum halepense* (L.) Pers.) and their influence on germination and seedlings growth. Beside control treatment, eight treatments for breaking dormancy of seeds were applied: seed immersion in distilled water for 24 hours; seed immersion in water at 60 °C for 1 hour; immersion in a 0.2% solution of potassium nitrate for 24 hours; immersion in 2% solution of sodium hypochlorite for 8 hours; treatments with concentrated sulphuric acid for 5 and 10 minutes; treatment with concentrated sulphuric acid for 5 minutes and germination in darkness; and combined treatment of sulphuric acid (5 minutes) and 1.5% potassium nitrate solution (2 hours). Average germination of Johnson grass seeds in control treatment was very low, only 2.8%. All dormancy breaking treatments increased seed germination and the highest germination was observed in treatment with combination of sulphuric acid and potassium nitrate (35.0%) and treatment with 2% sodium hypochlorite (30.6%). However, sodium hypochlorite reduced root length of seedlings for 40.9% compared to control, while all treatments had a positive effect on Johnson grass shoot length. Total seedlings length was the highest when seeds were treated for 5 minutes with sulphuric acid. None of the treatments showed significant effect on Johnson grass seedlings fresh weight. Seed germinated fastest in treatment with a combination of sulphuric acid and potassium nitrate (4.58 days), and slowest when seeds were immersed in water (8.16 days) and in the 2% solution of sodium hypochlorite (8.92 days).

Keywords: dormancy, germination dynamics, Johnson grass (*Sorghum halepense* (L.) Pers.), scarification

Sažetak

Cilj istraživanja bio je utvrditi učinkovitost različitih metoda prekida dormantnosti sjemena korovne vrste divlji sirak (*Sorghum halepense* (L.) Pers.) odnosno njihov utjecaj na klijavost i rast klijanaca. Uz kontrolni tretman, primijenjeno je osam tretmana za prekidanje dormantnosti sjemena: potapanje sjemena u destiliranu vodu tijekom 24 sata; zagrijavanje u vodi na 60 °C tijekom 1 sata; potapanje u 0,2% otopinu kalijevoga nitrata tijekom 24 sata; potapanje u 2% otopinu natrijevoga hipoklorita; tretiranje koncentriranom sumpornom kiselinom tijekom 5 i 10 minuta; tretiranje koncentriranom sumpornom kiselinom tijekom 5 minuta i naklijavanje u tami, te kombinirani tretman sumporne kiseline (5 minuta) i 1,5% otopine kalijevoga nitrata (2 sata). Prosječna klijavost sjemena divljeg sirka u kontrolnom tretmanu bila je vrlo niska, svega 2,8%. Svi tretmani prekidanja dormantnosti povećali su klijavost sjemena, a najviša klijavost zabilježena je u tretmanu s kombinacijom sumporne kiseline i kalijevoga nitrata (35,0%) te u tretmanu s 2% otopinom natrijevoga hipoklorita (30,6%). Međutim, primjena natrijevoga hipoklorita smanjila je duljinu korijena klijanaca divljeg sirka za 40,9% u odnosu na kontrolu, dok su svi tretmani imali pozitivan utjecaj na duljinu izdanka divljeg sirka. Ukupna duljina klijanaca bila je najviša kod tretiranja sjemena sumpornom kiselinom tijekom 5 minuta. Tretmani nisu pokazali značajan utjecaj na svježiu masu klijanaca divljeg sirka. Najbrže je klijalo sjeme u tretmanu s kombinacijom sumporne kiseline i kalijevoga nitrata (4,58 dana), a najsporije kod potapanja sjemena u vodu (8,16 dana) te u 2% otopinu natrijevoga hipoklorita (8,92 dana).

Ključne riječi: dinamika klijanja, divlji sirak (*Sorghum halepense* (L.) Pers.), dormantnost, skarifikacija

Introduction

Weed species possess outstanding characteristics such as adaptability, ability to reproduce large amount of seeds, resistance to unfavorable environmental conditions and germination periodicity (Šarić, 1991). The timing of germination is a critical moment in a plant's life (Donohue, 2005) and frequently depends on seed dormancy (Postma and Ågren, 2015). Seed dormancy represents incapacity of seed to germinate in a specified period of time under any combination of normal physical environmental factors that otherwise is favorable for its germination (Bewley, 1997). Dormancy can be classified as primary dormancy if it is induced during seed development resulting in seeds that are dormant when they are dispersed from the mother plant, or as secondary dormancy which is a result of unfavorable environmental conditions after seed dispersal (Vivian et al., 2008). The existence of a large population of weed seeds with varying degrees and states of dormancy enables weediness period to remain constant, continuously one year after another, and is the main reason why weeds reappear over a specific time period (Ali et al., 2012; Podrug et al., 2014). Number of established weed plants is strongly related to the portion of the seed bank that has been released from dormancy (Benech-Arnold et al., 2000). Knowledge on seed dormancy and weed emergence timing leads to better design of

an efficient weed management system (Dekker, 1999). Seed dormancy is also important when the post-emergence weed control tools are studied in laboratory and greenhouse experiments, where weed seed germination and seedling establishments are necessary (Mohammadi et al., 2013).

Johnson grass (*Sorghum halepense* (L.) Pers.) is a perennial species belonging to the family Poaceae. It is widespread weed in Europe, North and South America, Asia and Australia and New Zealand occurring on different habitats, in row crops, meadows and ruderal sites (Knežević, 2006; Nikolić et al., 2014). Johnson grass is a weed difficult to control since it produces extensively creeping rhizomes which regenerate easily and large number of seeds, up to 30 000 per plant (Warwick and Black, 1983). Majority of the Johnson grass seeds are highly dormant with up to 10% germination and seed dormancy is largely imposed by mechanical restriction of the seed coat which contains tannin compounds responsible for its reduced permeability to water (Bennet, 1973; Taylorson and McWorther, 1969). Mainly due to their dormancy, up to 60-70% of the seeds can remain viable after 25 years in the soil (Egley and Chandler, 1978).

Successful interruption of Johnson grass dormancy was recorded with various methods such as mechanical scarification and degluming (Krenchinski et al., 2015; Nosratti et al., 2012), chemical scarification with sulphuric acid sodium hypochlorite and hydrogen peroxide (Huang and Hsiao, 1987; Mohammadi et al., 2013; Podrug et al., 2014; Salimi and Termeh, 2002), and hot water treatments (Đikić et al., 2011), however, the results vary extremely, even among the same methods. According to Nosratti et al. (2012) seeds of various biotypes of Johnson grass respond differently to dormancy breaking treatments. So, the aim of the research was to evaluate different treatments for breaking seed dormancy of local population of Johnson grass and their effect on seed germination and seedlings growth.

Materials and methods

The experiment was conducted during 2014/2015 in Laboratory of Phytopharmacy at the Faculty of Agriculture in Osijek. Seeds of Johnson grass were collected in 2014 from Johnson grass plants growing on edges of maize fields in the Osijek-Baranja County. The collected seed were thoroughly cleaned and stored in paper bags until use. Prior to each experiment, the seeds were surface disinfected for 10 minutes in 1% solution of NaOCl and rinsed with distilled water (Siddiqui et al., 2009).

Treatments for breaking seed dormancy of Johnson grass seeds included: 1) control (untreated seeds); 2) immersion in distilled water for 24 hours; 3) immersion in hot water at 60 °C for 1 hour; 4) immersion in 0.2% solution of KNO₃ for 24 hours; 5) immersion in 2% solution of NaOCl for 8 hours; 6) concentrated H₂SO₄ for 5 minutes; 7) concentrated H₂SO₄ for 5 minutes and seed germinated in darkness; 8) concentrated H₂SO₄ for 10 minutes; 9) concentrated H₂SO₄ for 5 minutes plus immersion in 1.5% solution of KNO₃ for 2 hours.

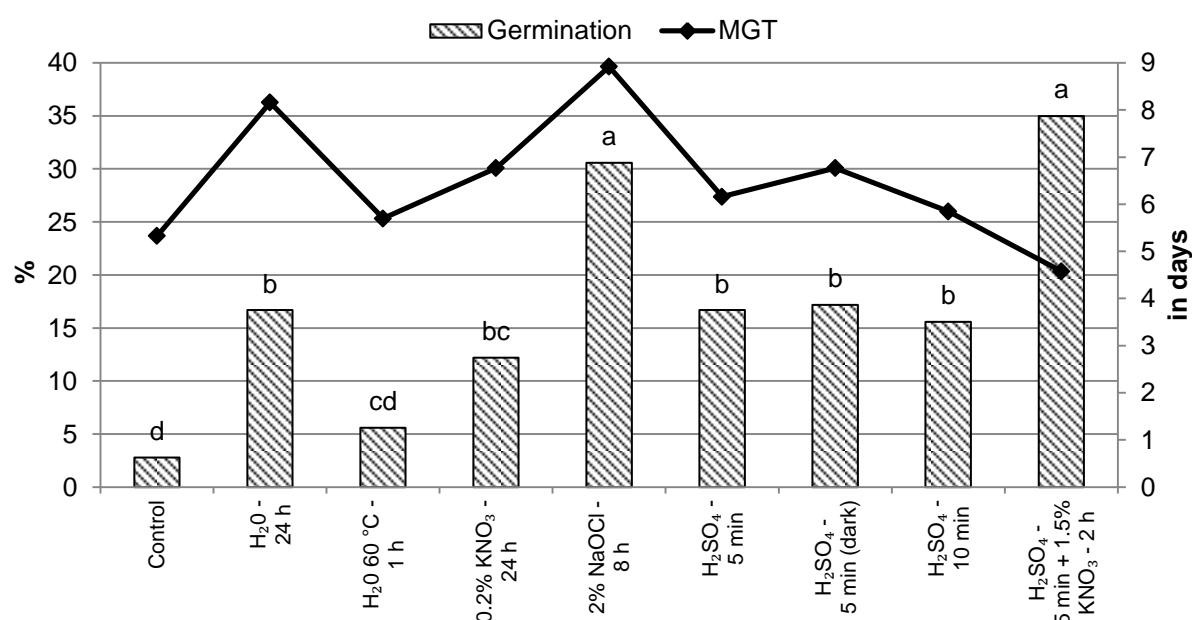
After application of different treatments, seeds of Johnson grass were germinated in Petri dishes. In each dish 30 weed seeds were placed on filter paper moistened with 5 ml of distilled water. In the seventh treatment Petri dishes were wrapped in aluminum foil in order for seeds to germinate in darkness. Seeds were germinated at

room temperature (20 ± 2 °C) on laboratory benches, while experiment lasted for 15 days.

Experiment was set up as completely randomized design; each treatment had three replications and experiment was repeated. Germinated seeds were recorded daily. Germination percentage was calculated for each replication using the formula: $G = (\text{Germinated seed} / \text{Total seed}) \times 100$. Mean germination time (MGT) was calculated according to the equation of Ellis and Roberts (1981): $MGT = \sum (Dn) / \sum n$, where n is the number of seeds that germinated on day D , and D is number of days counted from the beginning of germination. At the end of experiment seedling root length (cm), shoot length (cm) and fresh weight (mg) were determined. The collected data were analyzed statistically with ANOVA and differences between treatment means were compared using the LSD-test at probability level of 0.05.

Results

The effect of treatments on breaking dormancy of Johnson grass seed is presented in Figure 1. The lowest seed germination was recorded in the control and was 2.8%. Significant increase in germination was recorded in all treatments, except when seeds were immersed in hot water. The highest germination was observed with 2% solution of NaOCl and combined application of concentrated H_2SO_4 and 1.5% solution of KNO_3 and was 35.0 and 30.6%, respectively.



abc - means followed by the same letter within the column are not significantly different at $P < 0.05$.

Figure 1. Effect of dormancy breaking treatments on germination and mean germination time of Johnson grass seed

Grafikon 1. Utjecaj tretmana prekidanja dormantnosti na klijavost i prosječno vrijeme klijanja divljeg sirka

Mean germination time of Johnson grass seed differed among treatments, and compared to the control seed germinated faster only in treatment with concentrated H_2SO_4 and 1.5% solution of KNO_3 (Figure 1). In contrary, all other treatments prolonged seed mean germination time. Seeds germinated the slowest when immersed in distilled or NaOCl solution, for 2.83 and 3.59 days longer compared to the control.

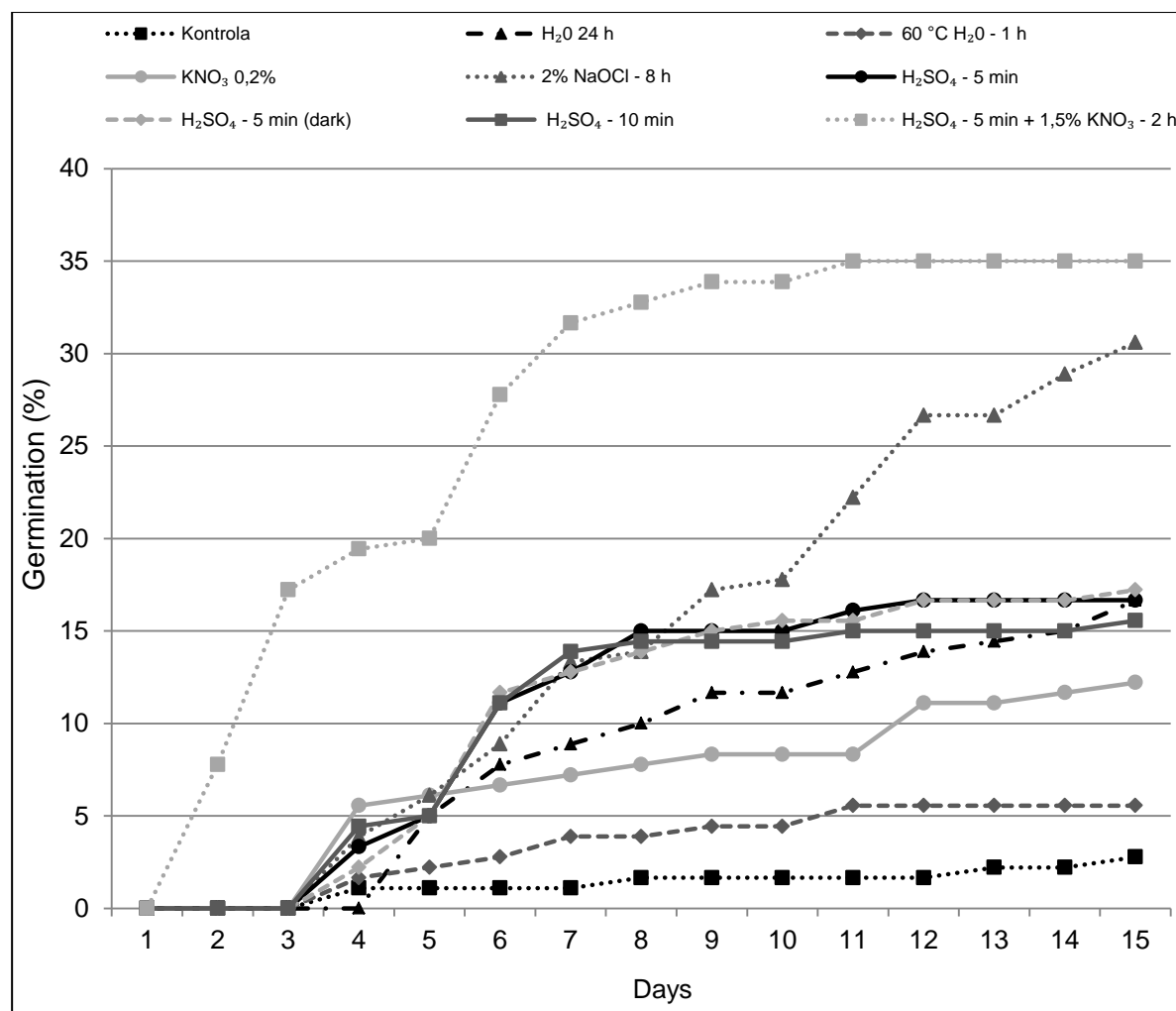


Figure 2. Effect of dormancy breaking treatments on seed germination dynamics of Johnson grass

Grafikon 2. Utjecaj tretmana prekida dormantnosti na dinamiku klijanja sjemena divljeg sirka

Germination dynamics of Johnson grass seed (Figure 2) indicated that in most treatments full germination was reached between 6 and 8 days after its onset. In treatment with a combination of concentrated H_2SO_4 and 1.5% solution of KNO_3 only 50% of the seeds germinated on the fifth day while full germination was recorded on the 11th day. In the treatment with NaOCl full germination was reached on the 15th day of the experiment.

Root and shoot length of Johnson grass seedlings differed significantly when dormancy breaking treatments were applied (Table 1). Combination of concentrated H_2SO_4 and 1.5% solution of KNO_3 increased seedling root length up to 31.8%, while NaOCl decreased root length for 38.6%. Shoot length of Johnson grass seedlings was increased in all treatments compared to the control, especially in treatment with concentrated H_2SO_4 for 5 minutes. Total seedlings length was significantly increased only in with concentrated H_2SO_4 for 5 minutes, for 50% compared to the control. None of the treatments significantly affected Johnson grass seedlings weight.

Table 1. Effect of dormancy breaking treatments on growth of Johnson grass seedlings

Tablica 1. Utjecaj tretmana prekida dormantnosti na rast klijanaca divljeg sirka

Treatment ^a	Root length (cm)	Shoot length (cm)	Total length (cm)	Seedlings weight (mg)
Control	4.4 b	5.8 e	10.2 bcd	26.7 ab
H ₂ O – 24 h	3.6 bc	8.4 bcd	11.7 bcd	25.9 ab
H ₂ O 60 °C – 1 h	3.8 bc	8.9 bc	12.8 ab	28.8 a
0.2% KNO ₃ – 24 h	3.3 bc	6.3 de	9.5 d	23.8 ab
2% NaOCl – 8 h	2.7 c	7.1 cde	9.8 cd	19.7 b
H ₂ SO ₄ – 5 min	4.3 b	11.0 a	15.3 a	24.2 ab
H ₂ SO ₄ – 5 min (dark)	3.3 bc	7.3 bcde	10.6 bcd	19.3 b
H ₂ SO ₄ – 10 min	3.4 bc	9.1 ab	12.5 bc	20.0 b
H ₂ SO ₄ – 5 min + 1.5% KNO ₃ – 2 h	5.8 a	6.7 de	12.5 bc	20.0 b

^a abc - means followed by the same letter within the column are not significantly different at $P < 0.05$.

Discussion

Germination of Johnson grass seeds in the control treatment amounted up to only 2.8% indicating that the large proportion of seeds was dormant. Application of dormancy breaking treatments resulted in the increase of germination, but the highest percentage of seeds germinated when treated with concentrated H_2SO_4 for 5 minutes and immersed in 1.5% KNO_3 solution for 2 hours (35.0%). Similarly, application of concentrated H_2SO_4 alone also increased seed germination but to a lesser extent, to 15.6 and 16.7%. Concentrated H_2SO_4 is used for breaking seed dormancy of many species with impermeable seed coat (Tigabu and Oden, 2001) since it causes modification or scarification of the hull or seed coat membranes, and also supplies the additional oxygen to the seed (Huang and Hsiao, 1987; Tao, 1982). Combination of concentrated H_2SO_4 and KNO_3 according to Shanmugavalli et al. (2007) increased germination of sorghum seed from 0 to 94%. Bewley and Black (1983) state that KNO_3 raises the ambient oxygen levels by making less oxygen available for citric acid cycle. In contrary, Mohammadi et al. (2013) reported that application of H_2SO_4 and KNO_3 had lesser affect oppose to treatments without KNO_3 ,

while according to Krenchinski et al. (2015) scarification with both diluted and concentrated H_2SO_4 had no effect on germination of Johnson grass.

Seed immersion in 2% solution of KNO_3 also affected seed dormancy of Johnson grass, however, the total germination was lower and was up to 12.2%. Contrary, results of both Đikić et al. (2011) and Podrug et al. (2014) suggested that KNO_3 had no significant effect on Johnson grass germination. Shanmugavalli et al. (2007) however stated that application of KNO_3 increased germination of sorghum seeds even more than H_2SO_4 , while Lemić et al. (2014) reported an increase in seed germination of common lambsquarter (*Chenopodium album* L.).

The immersion of seeds in 2% solution of NaOCl increased the seed germination to 30.6%. Other authors suggest the application of NaOCl as a successful method to overcome dormancy of Johnson grass (Mohammadi et al., 2013; Nosratti et al., 2012). The use of NaOCl increases the supply of oxygen in the seed embryo, but does not remove the seed hull (Hsiao and Quick, 1984).

The interruption of seed dormancy and total germination of 16.7% was observed in treatment with immersion of seeds in distilled water for 24 hours. Similar results in increase of Johnson grass germination were reported by Đikić et al. (2011) and Podrug et al. (2014), while according to Ravlić et al. (2015) this method also successfully overcomes the dormancy of redroot pigweed (*Amaranthus retroflexus* L.) seeds.

Hot water treatment failed to interrupt seed dormancy and confirms the results of other authors (Krenchinski et al., 2015; Podrug et al., 2014). Seed drying on the other hand on temperatures up to 90 °C showed effective in increasing after-ripening and germination of Johnson grass and barnyard grass (*Echinochloa crus-galli* (L.) PB.) (Huang and Hsiao, 1987; Salimi and Termeh, 2002).

A significant effect on seedlings growth was recorded, and NaOCl solution caused reduction in root length, while treatments generally promoted shoot length of Johnson grass. Growth inhibition of wild oat (*Avena fatua* L.) seedlings root length was also observed by Hsiao and Quick (1984) when Johnson grass seeds were immersed in NaOCl. Positive effects on seedlings length and weight were recorded with KNO_3 in redroot pigweed (Ravlić et al., 2015) and with gibberellic acid in sorghum (Shanmugavalli et al., 2007).

Mean germination time of Johnson grass seeds was the fastest in treatment with combination of concentrated H_2SO_4 and 1.5% KNO_3 solution, while other treatments prolonged germination compared to the control, especially immersion in NaOCl solution. According to Mohammadi et al. (2013) glume removal and treatment with concentrated H_2SO_4 increased number of germinated seeds per day.

Conclusions

The results of the study indicated that Johnson grass seed dormancy could be broken with various treatments, and both combination of H_2SO_4 and KNO_3 solution, and NaOCl solution proved as the best. However, application of NaOCl delayed seed germination and had inhibitory potential on seedlings root length.

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